

Laboratory Support of Food Borne Illness Investigations

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Laboratory Testing

- ✓ Data from laboratory tests are often key to effective public health investigation and control
- ✓ The adequacy and condition of the sample or specimen is of primary importance

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Outbreak Detection

- | | |
|-----------------------------------|--|
| ✓ Old Scenario | ✓ New Scenario |
| ○ Acute local outbreak | ○ Diffuse widespread outbreak |
| ○ High Dose/High Attack rate | ○ Low Dose/Low Attack rate |
| ○ Detected by group themselves | ○ Increase in "sporadic" cases |
| ○ Local Investigations | ○ Detected by lab based subtype surveillance |
| ○ Often local food handling error | ○ Complex multistate investigation |
| ○ Local Solution | ○ Industrial Contamination event |
| | ○ Industry wide implications |

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Objectives

At the end of this presentation, the participants will be able to:

- ✓ Discuss the various food testing that can be performed at the MTPHL
- ✓ Describe the steps necessary to ensure appropriate testing is performed
- ✓ List the organizations and networks involved in laboratory testing and food outbreak investigations

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Overview

- ✓ Criteria for Testing
- ✓ Collection of Clinical Specimens
- ✓ Key Items for a Food Sample Kit
- ✓ Collection of Food Samples
- ✓ Lab Requisition Forms and Chain of Custody
- ✓ Testing performed at the MTPHL
- ✓ Networks and Partners
- ✓ Scenario
- ✓ Contact Information

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Laboratory Support

- ✓ All food testing will be coordinated through the Montana Laboratory Services Bureau
 - Public Health Laboratory
 - Microbiology
 - Environmental Laboratory
 - Chemistry
- ✓ Contact us at 800-821-7284 (24/7)
- ✓ If we can't test – we will find a lab that can

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Criteria for Testing

- ✓ Food testing requires an epi-link
 - Shot gun approach doesn't work
 - Testing is targeted to specific agents
 - Agent implicated by food histories, onset of illness, symptoms
 - Ideally should have clinical specimens as well as food samples for comparison

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Criteria for Testing

- ✓ Communication and team work is the key for a successful outcome
- ✓ Environmental Health Assessment
- ✓ Epidemiological Investigation
- ✓ Laboratory Guidance and Support
- ✓ Education and Training
- ✓ Testing to be performed is determined by a foodborne outbreak investigation team

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Criteria for Testing (continued)

- ✓ Food Team will direct what samples are to be collected and submitted to the laboratory for testing
- ✓ Initial information may be insufficient for an adequate assessment
- ✓ Food Team will decide what samples to collect and then hold, either locally or at the MTPHL, until more information and assessment is available to direct testing
- ✓ Ensure that the opportunity to collect the appropriate specimens is not missed, but also utilizing costly and time consuming lab resources effectively

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Collection of Clinical Specimen

- ✓ Clinical specimens tested as early in the onset of illness as possible
- ✓ Suspect clinical **specimens** should be transported ASAP to a laboratory for testing
- ✓ Clinical **isolates** should be referred ASAP to the MTPHL
- ✓ Your local laboratory is trained to package and ship these specimens/isolates
- ✓ Courier service available in many areas of the state
- ✓ USPS if no courier service

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Food Sample Collection Kits

- ✓ Sterile Plastic Collection Containers
 - Whirl Paks –Various sizes
 - Small screw capped containers
- ✓ Sterile Disposable Collection Utensils
 - Spoons and Scoops
 - Tongs
 - Knife and cutting utensils
- ✓ Sterile Disposable Gloves
- ✓ Labels and Marker pens
- ✓ Chain of Custody Forms
- ✓ MTPHL Requisition Forms

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Collecting the Food Sample

- ✓ Wear sterile disposable gloves
- ✓ Use sterile technique
- ✓ Use sterile utensils for collection
- ✓ Use sterile transport containers
- ✓ Secure sample container –No leaking specimens
- ✓ Label all samples
- ✓ Description and condition of food sample
- ✓ Initials of sample collector
- ✓ Time and date of collection

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Collecting the Food Sample

- ✓ Recommend minimum 250 grams of solid food
 - Prefer 500 grams
- ✓ Recommend 250 minimum milliliters of liquid food
 - Prefer 500 milliliters
- ✓ In general, samples should be collected from the geometrical center of the food item, but should be representative of the food product
- ✓ If in doubt, send the entire food sample
- ✓ Commercially packaged food samples must be retained and transported in the original package
- ✓ Most food items are transported at refrigerator temperature
- ✓ If the food is already frozen at the time of collection, then transport the sample frozen

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Filling out the Requisition Form

- ✓ Working with your PH Nurse, use a form preprinted with county information
- ✓ Date and time of collection
- ✓ Physical description and condition of sample
- ✓ Site of collection
- ✓ Lot numbers
- ✓ Indicate Lab Tests to be performed, if known

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The image shows the MTPHL Requisition Form, a preprinted document with various sections for data entry. It includes a header for the Montana Department of Public Health & Human Services, a section for 'PATIENT INFORMATION' with checkboxes for various conditions, a section for 'TEST REQUESTS' with checkboxes for different laboratory tests, and a section for 'ADDITIONAL INFORMATION' with checkboxes for various sample types. The form is designed to be filled out by a healthcare provider or laboratory staff.

MTPHL Requisition Form

Preprinted with Location Information

Space for test requests

Space for additional information

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Chain of Custody

- ✓ Legal document verifying integrity of the sample from the moment of collection in the field until the final deposition of the sample
- ✓ Official Sample
 - Integrity of the sample is verifiable
 - Includes internal and external transfer information
- ✓ Unofficial Sample
 - Integrity of the sample is not verified
 - Example – leftovers from an individual's refrigerator
- ✓ 3 most important things when conducting testing for a food borne outbreak:
Document, Document and Document

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Chain of Custody Document for Credible Threats

The image shows a 'Chain of Custody Document for Credible Threats' form. It includes a 'Submitting Activity' section with checkboxes for credible threats and local health department contact. It has fields for 'Name and Title of Person Requesting Testing', 'Address', 'Contact Phone#', 'Location from Where Obtained', 'Description of Item to Be Tested', 'Date Collected', 'Name and Title of Person Collecting Article', and 'Time Collected'. At the bottom, there is a table for tracking the chain of custody with columns for 'Date & Time', 'Released By', 'Received By', and 'Purpose of Change in Custody'. The table has four rows for signatures and names.

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Food Samples

- ✓ Suspect foods should be stored refrigerated or frozen awaiting epidemiology investigation
 - Link between ingestion, clinical illness, suspected agent
 - Knowledge of intentional release
- ✓ If a food is implicated, transport to the PHL
 - Ship in cold condition
 - Styrofoam container with blue ice packs
- ✓ Courier service available in many areas of the state
- ✓ USPS if no courier service

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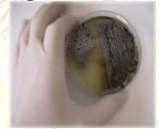
Testing Performed at MTPHL

- ✓ Bacterial Agents
 - *Salmonella*, *Shigella*, toxigenic *E. coli*, *Listeria*, *Vibrio*, *Campylobacter*
 - Agents of Bioterrorism (Tularemia, Brucellosis, Anthrax, Plague)
- ✓ Viral Agents
 - Norovirus (in process of validating for food)
- ✓ Toxins
 - *Staph* enterotoxin B
 - *C. botulinum*

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Testing for Bacterial Agents

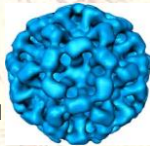
- ✓ Food Specimen Received for Bacterial Agents
 - Bacterial Enrichment
 - Culture and Identification
 - BAX amplification testing for *Salmonella*, *E. coli* O157, *Listeria*
 - Isolates subjected to DNA fingerprinting (PFGE)



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Testing for Viral Agents

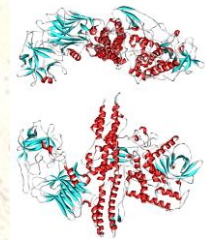
- ✓ Currently no validated method for Norovirus testing on food
 - Our FERN CoAg funded to try to validate a method
 - Real time PCR method
 - Isolates could be sequenced and uploaded to national database (CALICNET)



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Testing for Toxins

- ✓ *Staphylococcus* Enterotoxin A and B
 - M1M instrument
 - TRF
- ✓ *Clostridium botulinum*
 - M1M instrument
 - DIG-ELISA



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Networks and Partners

- ✓ Montana Department of Livestock
 - Regulatory Meat Program
- ✓ USDA
 - FSIS grant for validation of methods, capacity
- ✓ FDA
- ✓ FERN
 - Food Emergency Response Network
 - eLEXNET
- ✓ PulseNet
 - National database of DNA fingerprints of certain bacteria

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Department of Livestock

- ✓ MTPHL performs regulatory testing on meat samples under USDA FSIS guidelines
 - Ground meat
 - Carcasses
 - Processed meat
- ✓ *Salmonella*
- ✓ *E. coli* O157
- ✓ *Listeria monocytogenes*



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<http://www.fsis.usda.gov/index.asp>



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USDA - FSIS

- ✓ MTPHL has a continuing grant with FSIS maintain our capacity and to validate additional food methods and matrices
 - Validations of *Shigella spp.*, SEA, SEB, *C. botulinum* toxin, *E. coli* O157
- ✓ Food Defense Surveillance Assignments
 - USDA and FDA joint venture
 - Traced-back food products from retail to field
 - In 2007, FERN labs used for anthrax and *C. botulinum* toxin testing

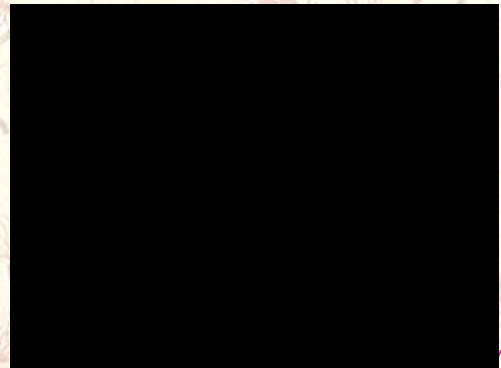
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FDA



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FDA – Anatomy of an Outbreak



Food Emergency Response Network

- ✓ FERN is a joint venture of USDA, FDA, CDC, EPA
- ✓ Network of state and federal laboratories that are committed to analyzing food samples in the event of a biological, chemical, or radiological terrorist attack
- ✓ Coordinates the sample distribution and nationwide surge capacity of the laboratories
- ✓ Facilitates secure communication and data exchange between laboratories (eLEXNET)
- ✓ Confirms the safety of the food supply using analytical tests

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FERN Public Web Page



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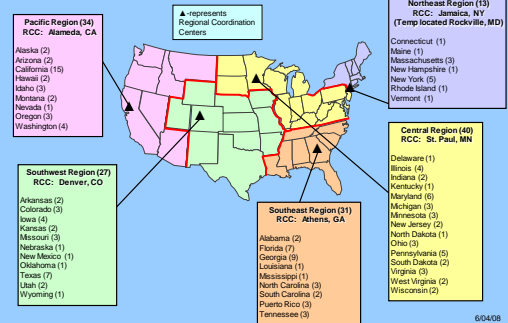
FERN

Critical roles related to food security and food defense include:

- ✓ **Prevention**
 - FERN provides a national surveillance program that will offer early means of detecting threat agents in the American food supply
- ✓ **Preparedness**
 - FERN prepares the nation's laboratories to be able to respond to food-related emergencies
- ✓ **Response**
 - FERN offers significant surge capacity that will strengthen the nation's response towards widespread complex emergencies, intentional or inadvertent related to agents in food
- ✓ **Recovery**
 - The FERN network of laboratories enhances the ability of the country to restore confidence in the food supply following a threat or an actual emergency targeting the nation's food supply.

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Food Emergency Response Network Federal, State, and Local Laboratories = 138 Total



6/04/08

eLEXNET – Electronic Laboratory Exchange Network

- ✓ FERN uses eLEXNET for Collaboration & Data Sharing:
 - FERN National Program Offices share current information, meeting minutes, documentation, and guidance information
 - FERN Subcommittees conduct discussions, disseminate information, and develop documentation
 - FERN Methods Repository on eLEXNET
 - 113 Lab Participants can review information on samples that have been submitted by eLEXNET's participating laboratories
 - FERN Proficiency & Surveillance Assignments: FERN related data can be captured and reported independently

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Recent FERN Activities

- ✓ Spinach outbreak - EHEC
- ✓ Pet food contamination – Melamine
- ✓ Tomatoes/Peppers – *Salmonella*
- ✓ Peanut Butter – *Salmonella*
- ✓ Food defense assignments

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PulseNet

- ✓ Standardized PFGE methods for DNA fingerprinting of disease-causing bacteria
 - Current gold standard for bacterial DNA fingerprinting
 - Sensitive, specific and reproducible
- ✓ Patterns generated and entered into an electronic database at MTPHL
- ✓ Patterns uploaded to national CDC database
- ✓ Database managers at CDC perform regular searches looking for clusters
 - "Fishing" for related cases in a "sea of infections"
- ✓ Clinical Laboratory Specialists perform regular searches on local databases looking for clusters

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Pulsed Field Gel Electrophoresis

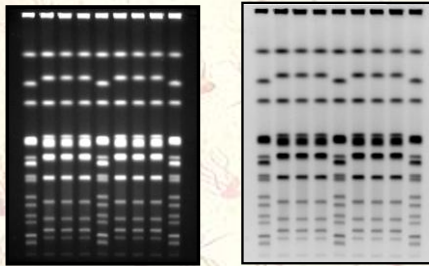


**Bacterial DNA
Fingerprinting**

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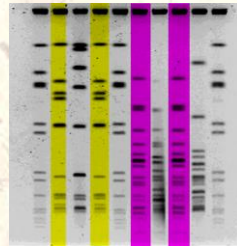
Imaged PFGE Gel

Example: *Salmonella enteritidis*



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PFGE Patterns: Epidemiologically Linked Patients



- ✓ Listeria PFGE patterns for patients (#1 & #2) were indistinguishable possibly indicating a common source of infection
- ✓ Asc-I patterns: highlighted in yellow
- ✓ Apa-I patterns: highlighted in purple

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PulseNet

- ✓ Plays a vital role in surveillance and investigation of food-borne illness outbreaks
- ✓ Can find outbreaks that are geographically far apart
- ✓ Distinguishes outbreak cases from concurrent sporadic cases

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PulseNet History

- ✓ 1993 – outbreak of *E. coli* O157 in western United States
- ✓ CDC scientists did PFGE fingerprinting
 - Strain found in patients had the same pattern as strain found in hamburger patties
- ✓ Prompt recognition prevented additional illnesses
- ✓ CDC deployed the technology to state and local laboratories

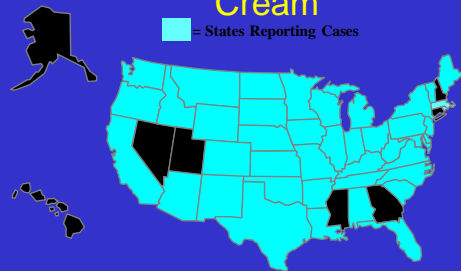
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Schwan's Ice-Cream Associated Outbreak, 1995



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States Reporting Cases Associated with Schwan's Ice Cream



Size of Outbreak: approximately 250,000 cases in the U.S.

E. coli O157:H7 Outbreak from Fresh Spinach, 2006



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Salmonella tennessee

PFGE-XbaI



AL-8002774-06

MT2007603021

0611ALJNX-1c (JNXX01.001)

- ✓ Montana had two matching cases
 - 85 year old female in Yellowstone Co, Nov 2006
 - 63 year old male in Silver Bow Co, Jan 2007

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S. tennessee Outbreak

- ✓ Epidemiology Investigation of Jan. patient
 - Inpatient at hospital at time of diarrhea
 - Source of Salmonella was unknown
 - After PulseNet match, asked about peanut butter consumption
 - Wife had been bringing patient peanut butter and jelly sandwiches from home
 - Opened jar had product code 2111, with the production dates in the "window period"

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Food Borne Illness Investigation Scenario



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Scenario (1)

- ✓ Physician calls the local health department
 - Has just seen a husband and wife
 - Both are sick with a diarrheal illness
 - Watery diarrhea, some blood in the stool, abdominal cramping, dehydration
 - Report that they both ate breakfast at a local restaurant and became ill shortly after
- ✓ There have been no other reports of similar illness to the health department

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Scenario (2)

- ✓ Cultures were submitted to the local laboratory
- ✓ After 2 days, the cultures on both patients have grown *E. coli* O157

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Scenario (3)

- ✓ Food Team is assembled
 - Epidemiology investigation is initiated
 - Questionnaire administered to husband and wife
 - Environmental Health Risk assessed
 - Not just the food they eat
 - Where they ate – possible restaurant inspection
 - What other risk factors are present?
- ✓ Since laboratory reports *E. coli* O157, interested in the 1 – 3 days prior to onset of illness

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Scenario (4)

- ✓ Investigation implicates food products
 - 3 days prior to onset:
 - BBQ'd Ground beef patties, eaten with
 - Sprouts
 - Lettuce
 - Tomato
 - Onion
 - Cheese
 - Mayonnaise
 - Ketchup
 - Pickles
- ✓ Food samples are collected

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Scenario (5)

- ✓ Ground Beef submitted for testing
- ✓ Sprouts, Lettuce, Tomato collected, but held at refrigerator temperature for results of the ground beef testing
- ✓ *E. coli* O157 is recovered from the ground beef
- ✓ DNA Fingerprinting is performed on both isolate from patients and isolate from ground beef

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Scenario (6)

- ✓ DNA fingerprint is uploaded to national PulseNet database
- ✓ A match is found in Colorado
- ✓ One week later, there are 10 matches in the PulseNet database from 4 other states
- ✓ Implicated ground beef had been distributed to 10 western states and is being recalled



ANOTHER SUCCESS STORY

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MTPHL Support

- ✓ Will advise on proper collection, storage and transport
- ✓ Laboratory works with the Food Team to track clinical specimens and food samples
- ✓ Performs testing to confirm diagnosis
- ✓ Performs testing to find relatedness of strains to the outbreak strain
- ✓ Point of contact for all laboratory issues
- ✓ 800-821-7284 (24/7)

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Questions?

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